

From Granaries to Insectaries

NIR Technology Helps Human Health

It all started at a 1998 meeting of entomologists in Gainesville, Florida. James Throne, an expert on stored-products insects with the Agricultural Research Service, stood at the podium sharing his lab's latest research on detecting tiny insects that lurk inside grains. By using an automated single-kernel handling system coupled with a near-infrared spectrometer, he and colleagues found they could pinpoint grain kernels that looked ordinary on the outside but harbored growing insects inside.

Many in the audience tuned in to hear more. Insect-infested grains are a problem for the milling and processing industries—and for consumers who typically cringe at the sight of a weevil in their bag of flour or box of cornflakes.

The ARS technology relies partly on near-infrared (NIR) light, that part of the light spectrum that's invisible to the human eye. Each living organism absorbs NIR radiation uniquely, depending on its particular chemistry. The energy reflected back can be analyzed by a spectrometer that researchers then interpret.

James Baker, an ARS entomologist and colleague of Throne's, first thought to use NIR spectroscopy for singling out bug-infested kernels.

"Insects inside of grains are one of the hardest defects to detect," he says. "For instance, a female grain weevil will bore into a wheat kernel to lay an egg. She'll seal the opening with a tiny, gelatinous plug. The mark left behind is hard to see with the naked eye."

NIR spectroscopy seemed a natural tool for tackling this job, since its light zeroes in on molecules made up of

nitrogen, carbon, hydrogen, and oxygen within biological materials. Grains and insects have different amounts and combinations of these molecules.

"We knew NIR spectroscopy was useful for analyzing the protein inside grains, so I wondered if it couldn't detect developing insects, too," Baker says. He

of the Division of Parasitic Diseases at the Centers for Disease Control and Prevention (CDC) in Atlanta, Georgia, had his sights on another pest—one that leaves a more direct mark on human health and well-being.

Identifying Malaria Carriers

Wirtz wondered if the NIR-based technology could help identify mosquitoes infected with the malaria parasite.

"Researchers used to have to dissect the insects," he says, "and remove and analyze the salivary glands to see whether they were infected."

The ARS technology could possibly speed this critical identification process. He thought it might also be able to distinguish between different species of the genus *Anopheles* that carry the malaria-causing parasite.

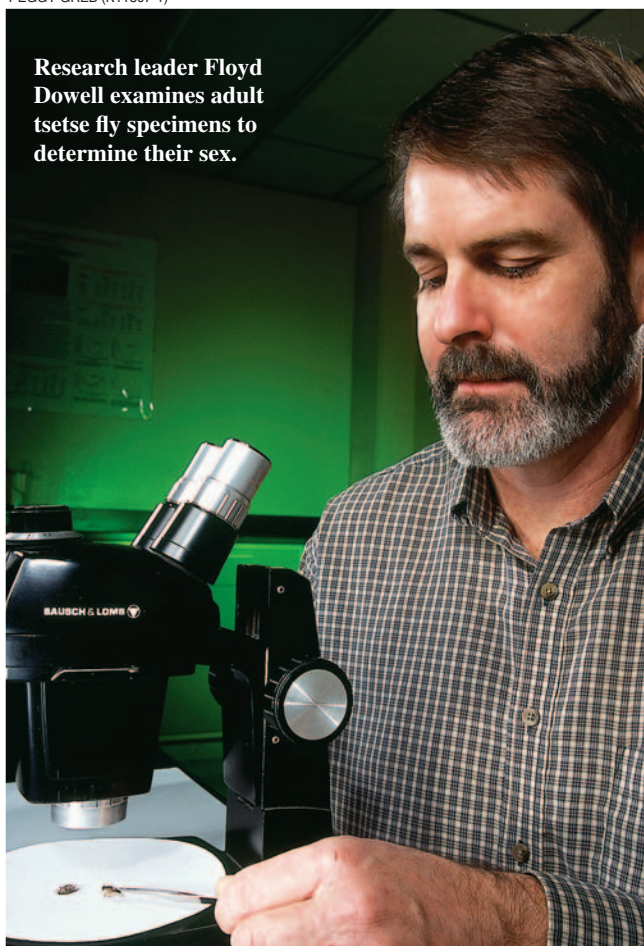
"There are species of *Anopheles* mosquitoes with different levels of significance in disease transmission," Wirtz explains. "We hoped the NIR technology could tell apart these look-alike species."

According to CDC entomologist Mark Benedict, another challenge is determining how old the mosquitoes are, since age plays an important role in disease transmission. "It takes time for mosquitoes to become infected," he says. "With older populations, there's more potential for transmission."

Floyd Dowell, ARS engineer and chief architect of the NIR sorting instrument, was able to find extra time to pursue this unexpected offshoot of his research. But now he would be working with living, breathing insects—instead of the usual, easy-to-handle kernels of grain.

PEGGY GREB (K11807-1)

Research leader Floyd Dowell examines adult tsetse fly specimens to determine their sex.



and Throne both work at the ARS Grain Marketing and Production Research Center in Manhattan, Kansas.

Back at the meeting, someone else—with a different set of research objectives—was listening to Throne's talk. But his interest wasn't in grains or their larval stowaways. Robert Wirtz, who heads the Entomological Branch



Female tsetse fly specimen
(*Glossina* spp.).

“Since adult insects are about the same size as kernels, we didn’t have to change the design settings used by the instrument for measuring grains,” he says. “But we did have to hand-place the insects into it.” Then the instrument accurately identified the species of mosquito in just seconds.

Attempts to identify malaria-infected mosquitoes weren’t as successful, but Wirtz says he hasn’t given up on that possibility yet.

“Our goal,” he says, “is to use the technology being developed by ARS to someday determine the species, age, and infection status of mosquitoes. This would be a huge benefit for controlling malaria, West Nile virus, and other diseases transmitted by mosquitoes.”

Energized by the mosquito work, Benedict and Wirtz wanted to see whether NIR could help distinguish between male and female pupae of the tsetse fly—an insect wreaking havoc in 37 countries in Africa. Coincidentally, a tsetse fly pupa is about the same size as a grain kernel and could be fed automatically through the NIR sorting system.

The tsetse fly carries the trypanosome parasite, which causes sleeping sickness in humans and nagana in livestock. Without medical treatment, sleeping sickness—or trypanosomiasis—is always fatal. According to the World Health Organization, more than 60 million people, mainly in rural and agricultural areas of afflicted African regions, are at risk.

Benedict thought NIR spectroscopy could help refine the sterile insect technique (SIT) that he was helping researchers with the United Nations Food and Agriculture Organization (FAO) and the International Atomic Energy Agency (IAEA) pursue to suppress tsetse fly numbers in ravaged parts of Africa.

From Screwworms to Tsetse Flies

SIT was pioneered by another ARS entomologist, the late Edward F. Knippling,

in the 1950s as an alternative to conventional pesticides. Often described as “birth control for insects,” this approach knocks down problem insect populations by releasing large numbers of sterilized males, which behave and mate like other males, but aren’t able to fertilize eggs. Over time and with continual releases, the targeted insect population dwindles and ultimately crashes. The technique was first used to rid North America of livestock-parasitizing screwworm flies.

But the SIT for tsetse needed a more rapid way to separate male and female flies. Males must be pulled aside to be irradiated, and females are needed to spawn future generations. Researchers also want to avoid accidentally mixing females with the males to be released, since females have a much greater potential to carry disease.

“Initially, when telling the sexes apart, we had to look at every adult by hand after it emerged,” says Benedict. “It was very time-consuming.”

To gain access to thousands of tsetse fly pupae, Dowell traveled to Seibersdorf, Austria, in January 2004 to the FAO-IAEA Agriculture and Biotechnology Laboratory. He and the others wanted to see how soon in the insect’s development NIR could distinguish between males and females. The earlier the better, as researchers need ample time to irradiate the males and transport them to strategic release sites.

“We thought that we could tell them apart at 1 day before their emergence as adults—but probably not at 20 days before emergence, when the pupae are less developed,” Dowell says.

As it turned out, they could sex them best at 5 days before the pupae emerge as adults. At that point, the differences in reflected light, or spectra, of female and male pupae were most apparent.

“These differences might be explained by changes in cuticle thickness or by



Research leader James Throne and engineer Elizabeth Maghirang place tsetse fly pupae in an automated scanning and sorting system.

ovarian development in the females,” Dowell speculates. “We can show without a doubt that there are differences between males and females, but unfortunately we lack the science to explain why. This will be a topic of future research if we can continue working in this area.”

According to ARS scientists, the NIR technology may work better for tsetse fly sex separation than for its original, intended use in grains. FAO-IAEA has purchased an automated NIR sorting instrument and will begin routinely sorting tsetse fly pupae in 2005.

“This is exciting technology and has lots of potential impact in the grain industry and the field of entomology—in addition to the potential human impact, particularly in underdeveloped countries,” says Dowell.—By **Erin Peabody**, ARS.

This research is part of Quality and Utilization of Agricultural Products (#306) and Veterinary, Medical, and Urban Entomology (#104), two ARS National Programs described on the World Wide Web at www.nps.ars.usda.gov.

To reach scientists featured in this article, contact Erin Peabody, USDA-ARS Information Staff, 5601 Sunnyside Ave., Beltsville, MD 20705-5129; phone (301) 504-1624, fax (301) 504-1641, e-mail ekpeabody@ars.usda.gov. ♦